

Potential for regulatory genetic networks of gene expression near a stable point

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A description for regulatory genetic network based on generalized potential energy is constructed. The potential energy is derived from the steady state solution of linearized Fokker-Planck equation, and the result is shown to be equivalent to the system of coupled oscillators. The correspondence between the quantities from the mechanical picture and the steady-state fluctuations is established. Explicit calculation is given for auto-regulatory networks in which, the force constant associated with the degree of protein is very weak. Negative feedback not only suppresses the fluctuations but also increases the steepness of the potential. The results for the fluctuations agree completely with those obtained from linear noise Fokker-Planck equation.

A regulatory network of gene expressions consists of a group of genes which co-regulate one another's expressions. Such networks provide a fundamental description of cellular function at the DNA level. Recently, the advance of experimental techniques in constructing synthetic networks with the ability of monitoring them has provided some essential elements, such as switch[1, 2, 3] and oscillator[4, 5], for the design of biological circuits. In modeling the dynamics of a regulatory network, rate-equation approach is often used; the approach reflects the macroscopic observation with deterministic nature. However for systems with small molecular number, intrinsic fluctuations become important. The noise-induced effect may be incorporated into the framework by employing the master equation and then proceeding via stochastic Monte Carlo simulations. In general, master equation is discrete in nature. By using the technique of Ω -expansion[6], we may convert master equation to continuous Fokker-Planck equation which, then, is managed analytically by various approximations. Significant progress has been made along this line in understanding the regulation mechanism. One of the noticeable examples is the auto-regulatory networks of a single gene for which, the protein, encoded in the gene, serves as the regulator of itself through either negative or positive feedback. Such autoregulation is a ubiquitous motif in biochemical pathways. It was demonstrated by Becskei and Serrano that an autoregulatory network with negative feedback may gain stability[7]. Further analyses was given by Thattai and van Oudenaarden[8] and by Ozbudak et al.[9], and the results indicate that noise is essentially determined at the translational level and negative feedback can suppress the intrinsic noise. Moreover, Tao and Tao et al. used the linear noise Fokker-Planck equation to study the fluctuations and obtained the results consistent qualitatively with previous works[10, 11].

One may conclude from the results above that the intrinsic noise associated with a genetic network is closely related to its regulation scheme. This Letter then attempts to provide a physical picture on this relation via the establishment of a mechanical analogous system. To achieve this, we first construct the solution of non-equilibrium steady state for the Fokker-Planck equation near a stable point. Then, the potential of the system, defined as the negative of the logarithm of the solution, can be first approximated as an harmonic oscillator potential. Subsequently, we introduce a measure for the steepness of the potential near a stable point and give the exact relations between the force constants of coupled oscillators and the correlations of fluctuations. Thus, the physical property of a regulation scheme can be revealed from the corresponding mechanical analogue specified by the force constants of coupled oscillators. This paper starts with the general construction for a d -dimensional regulation network, followed by the explicit calculations of auto-regulatory networks.

Consider a d -dimensional regulatory network of gene expression. The network is specified by the macroscopic rate equations

$$\dot{x}_i = f_i(x) \quad (1)$$

with $i = 1, 2, \dots, d$, and the drift force f_i defined as

$$f_i(x) = R_i(x) - \phi_i x_i. \quad (2)$$

Here, $x^\tau = (x_1, x_2, \dots, x_d)$ with the superscript τ for the transpose of a vector, x_i represents the concentration of mRNA or protein, the function $R_i(x)$ describes the synthesis or feedback regulation of molecule i , and the constant ϕ_i denotes the degradation rates of x_i . The network is assumed to form a chain with the nearest neighboring regulation, $R_i(x) = R_i(x_{i-1}, x_{i+1})$; however, the formulation presented in this work can be extended to more complicated cases

straightforwardly. The fluctuation may be incorporated into Eq. (1) by means of the master equation approach. For this, we introduce the volume factor Ω to give the molecular number $n^\tau = (n_1, n_2, \dots, n_d)$ as $n^\tau = \Omega x^\tau$. In terms of molecular numbers n , the corresponding master equation of Eq. (1) can be written as

$$\frac{\partial P(n, t)}{\partial t} = \sum_{i=1}^d (E_{i+} - 1) [(\phi_i n_i) P(n, t)] + \Omega \sum_{i=1}^d R_i(x) [E_{i-} - 1] P(n, t), \quad (3)$$

where $P(n, t)$ is the probability distribution, and the step operators $E_{i\pm}$ are defined as

$$E_{i\pm} G(n_i) = G(n_i \pm 1) \quad (4)$$

for a function of molecular numbers $G(n)$. Then, the technique of Ω -expansion[6] is employed to transfer the discrete process of Eq. (3) to a continuous process described by the Fokker-Planck equation,

$$\frac{\partial \rho(x, t)}{\partial t} + \nabla \cdot J(x, t) = 0, \quad (5)$$

where $(\nabla)^\tau = (\partial/\partial x_1, \partial/\partial x_2, \dots, \partial/\partial x_d)$, $\rho(x, t)$ is the distribution density, $J(x, t)$ is the density current defined as

$$J(x, t) = f(x) \rho(x, t) - \frac{1}{\Omega} [D(x) \cdot \partial] \rho(x, t), \quad (6)$$

and the elements of the diffusion matrix $D(x)$ are

$$D_{ij}(x) = \delta_{i,j} \left[\frac{R_i(x) + \phi_i x_i}{2} \right] \quad (7)$$

with the Kronecker delta $\delta_{i,j} = 1$ for $i = j$ otherwise 0, note we do not sum over repeated indices.

We are interested in the behavior of $\rho(x, t)$ for the region near a equilibrium stable point of Eq. (1), say x^* . After expanding the density current $J(x, t)$ of Eq. (6) around the stable point, we obtain the linearized Fokker-Planck equation for the new variable $y = x - x^*$ as

$$\frac{\partial \rho_L(y, t)}{\partial t} + \nabla \cdot J_L(y, t) = 0, \quad (8)$$

where $J_L(y, t)$, which contains only the leading order ($1/\Omega$) of $J(x, t)$, is

$$J_L(y, t) = \left[F(x^*) \cdot y - \frac{1}{\Omega} D(x^*) \cdot \nabla \right] \rho_L(y, t), \quad (9)$$

with the force matrix $F(x^*)$ defined as $F_{ij}(x^*) = \partial f_i(x) / \partial x_j|_{x=x^*}$, and noting the fact that y itself is of order $\frac{1}{\omega}$. This leads to a Ornstein-Uhlenbeck process in which, the drift force is linear and the diffusion is given by a constant matrix[6, 12]. The stationary solution of Eq. (8), characterized by the condition $\nabla \cdot J_L^S(y) = 0$, can be expressed as

$$\rho_L^S(y) = \frac{1}{Z} \exp[-\Phi(y)] \quad (10)$$

with

$$Z = \int_{-\infty}^{\infty} \cdots \int_{-\infty}^{\infty} \left(\prod_{m=1}^d dy_m \right) \exp[-\Phi(y)] \quad (11)$$

and

$$\Phi(y) = \frac{1}{2} y^\tau \cdot U(x^*) \cdot y, \quad (12)$$

where Z can be referred as the partition function, and U is a real symmetric $d \times d$ matrix[12]. Note that the temperature in this work is always set to be 1, $k_B T = 1$, and hereafter we drop the arguments for all matrix elements known to be functions of the equilibrium stable point x^* . One can determine the matrix U by substituting Eq. (10) directly into the condition $\nabla \cdot J_L^S(y) = 0$ to obtain

$$\text{tr}(F + \frac{1}{\Omega} D \cdot U) - y^\tau \cdot (U \cdot F + \frac{1}{\Omega} U \cdot D \cdot U) \cdot y = 0. \quad (13)$$

To solve this for the matrix U , we follow an elegant method proposed by Ao[13] and Kwon et al.[14] to factorize the force matrix as

$$F = -\frac{1}{\Omega} [D + Q] \cdot U, \quad (14)$$

where D is the symmetric diffusion matrix given by Eq. (7), and Q is an antisymmetric matrix which has to be determined. Such factorization amounts to decomposing the density current into two parts, $J_L^S(y) = j_d^S(y) + j_c^S(y)$. The first term of Eq. (14) corresponds to the dissipative part which generates a motion towards the origin with vanishing density current, $j_d^S(y) = 0$; meanwhile, the second term is the cyclic part with a divergence-free current density, $j_c^S(y) = -(1/\Omega) Q \cdot U \cdot y \rho_L^S(y)$, which generates a circulating motion around the constant surface of $\Phi(y)$. By substituting Eq. (14) into Eq. (13), we obtain the relation

$$F \cdot Q + Q \cdot F^\tau = F \cdot D - D \cdot F^\tau, \quad (15)$$

which gives enough conditions to determine the matrix Q completely. Thus, the function $\Phi(y)$ of Eq. (12), which can be referred as the potential energy for the system near a stable point x^* , becomes

$$\Phi(y) = -\frac{\Omega}{2} y^\tau \cdot \left[(D + Q)^{-1} \cdot F \right] \cdot y. \quad (16)$$

A more intuitive physical picture about the characteristics of the system may be revealed by mapping $\Phi(y)$ to the potential energy of the system of coupled oscillators,

$$\Phi(y) = \frac{\Omega}{2} \left[\sum_{i=1}^d \kappa_i y_i^2 + \sum_{i>j} \kappa_{ij}^c (y_i - y_j)^2 \right], \quad (17)$$

which can be casted in the form of

$$\Phi(y) = \frac{\Omega}{2} y^\tau \cdot V(\kappa, \kappa^c) \cdot y. \quad (18)$$

Note that though the regulations of the network only come from the nearest neighbors, the couplings of oscillators may not be restricted to the nearest neighbors. The force constants, κ and κ^c , can be specified by equating Eq. (18) to Eq. (16), and the characteristics of the network near a stable point can be expressed in terms of the force constants. Firstly, based on the partition function of Eq. (11), which is reduced to

$$Z = \left(\frac{2\pi}{\Omega} \right)^{d/2} [\det V(\kappa, \kappa^c)]^{-1/2} \quad (19)$$

with $\det V(\kappa, \kappa^c)$ for the determinant of the matrix $V(\kappa, \kappa^c)$ of Eq. (18), we may introduce the effective free energy difference, $\Delta G = -\ln Z$, to describe qualitatively the steepness of the potential. A stable point with larger ΔG value is more easy to focus with less fluctuations. Furthermore, the variances and covariance of x_1 and x_2 , defined as $\sigma_{i,j}^2 = \langle x_i x_j \rangle - x_i^* x_j^*$, can be evaluated by using the distribution $\rho_L^S(y)$,

$$\sigma_{i,j}^2 = \frac{1}{Z} \int_{-\infty}^{\infty} \cdots \int_{-\infty}^{\infty} \left(\prod_{m=1}^d dy_m \right) y_i y_j \exp \left[-\frac{\Omega}{2} y^\tau \cdot V(\kappa, \kappa^c) \cdot y \right]. \quad (20)$$

The formulation is applied to two-dimensional regulatory networks, and the results are given explicitly in the followings.

Consider the case of $d = 2$ with regulation functions $R_1(x_2)$ and $R_2(x_1)$. The force matrix is

$$F = \begin{pmatrix} -\phi_1 & r_1 \\ r_2 & -\phi_2 \end{pmatrix}, \quad (21)$$

where r_1 and r_2 are defined as $r_1 = \partial R_1(x_2) / \partial x_2|_{x_2=x_2^*}$ and $r_2 = \partial R_2(x_1) / \partial x_1|_{x_1=x_1^*}$. Then, the antisymmetric matrix Q , determined by Eq. (15), is

$$Q = \begin{pmatrix} 0 & W \\ -W & 0 \end{pmatrix} \quad (22)$$

with $W = [r_2 D_{11} - r_1 D_{22}] / (\phi_1 + \phi_2)$. The F and Q matrices given above with the D matrix of Eq. (7) yield the potential energy of Eq. (17) as

$$\Phi(y) = \frac{\Omega}{2} \left[\kappa_1 y_1^2 + \kappa_2 y_2^2 + \kappa_{1,2}^c (y_1 - y_2)^2 \right], \quad (23)$$

where the force constants are $\kappa_1 = [-r_2 D_{11} + (2\phi_1 - r_1) D_{22} + (2r_2 - \phi_2 + \phi_1) W] / 2 (D_{11} D_{22} + W^2)$, $\kappa_2 = [(2\phi_2 - r_2) D_{11} - r_1 D_{22} - (2r_1 + \phi_2 - \phi_1) W] / 2 (D_{11} D_{22} + W^2)$, and $\kappa_{1,2}^c = [r_1 D_{22} + r_2 D_{11} + (\phi_2 - \phi_1) W] / 2 (D_{11} D_{22} + W^2)$. For the effective free energy difference, we rescale ΔG by adding a volume factor, $\Delta \bar{G} = -\ln Z - \ln [\Omega / (2\pi)]$; then, $\Delta \bar{G}$ becomes half the logarithm of $\det V(\kappa, \kappa^c)$, and it is $\Delta \bar{G} = (\frac{1}{2}) \ln [\kappa_1 \kappa_2 + (\kappa_1 + \kappa_2) \kappa_{1,2}^c]$. Moreover, for the potential energy of Eq. (23) the variances and covariance of Eq. (20) become $\sigma_{1,1}^2 = [(\kappa_2 + \kappa_{1,2}^c) / \Omega] \exp(-2\Delta \bar{G})$, $\sigma_{2,2}^2 = [(\kappa_1 + \kappa_{1,2}^c) / \Omega] \exp(-2\Delta \bar{G})$, and $\sigma_{1,2}^2 = (\kappa_{1,2}^c / \Omega) \exp(-2\Delta \bar{G})$. Thus, expressed in terms of mechanical quantities we summarize the features of the system implied by the potential energy as follows. In general, the $\Delta \bar{G}$ value characterizes the global steepness of the quadratic potential; the increase of the $\Delta \bar{G}$ value makes the potential more sharper and, hence, reduces the fluctuations. However, for the same $\Delta \bar{G}$ value the details of potential shape may have an effect on the variances and covariance of components; the variance of one component is proportional to the force constant of the other and to the coupling strength between the two, and the covariance between the two is proportional to the coupling strength.

We apply the above results to study the regulation of an auto-regulatory network of a single gene, which describes the central dogma of gene expression, transcription and translation. The two variables, x_1 and x_2 , refer to the concentrations of mRNA and protein, respectively. In this study, we use the most common noise-attenuating regulatory mechanism, called negative feedback and described by Hill function $R_1(x_2) = k_{\max} / [1 + (x_2/k_d)^\beta]$. Here, k_{\max} is the maximum transcription rate of mRNA, k_d is the binding constant specifying the threshold protein concentration at which the transcription rate is half its maximum value, and β is the Hill coefficient. On the other hand, we set $R_2(x_1) = k_2 x_1$, where k_2 is the translation rate of protein. Then, a stable equilibrium, x^* , is characterized by two conditions: $\phi_1 \phi_2 - r_1 (x_2^*) k_2 > 0$ and $\phi_1 + \phi_2 > 0$. Subsequently, one can use Bendixson's criterion to further conclude that there is no any cycles, only one equilibrium exists[15]. For the values of the parameters, we mainly follow those given in Refs. [8, 11]. The half-lives of mRNA molecules and proteins are set as 2 minutes and 1 hour, respectively; this leads to $\phi_1 = (\ln 2) / 2$ and $\phi_2 = (\ln 2) / 60$ in the unit of $(\text{min})^{-1}$. The average size of a burst of proteins, $b = k_2 / \phi_1$, is set as 10, this leads to $k_2 = 5 (\ln 2)$. By using the fact that the protein concentration is about 1200 when $\beta = 0$ (no feedback), we set $k_{\max} = 3[16]$. To study the effect of the strength of negative feedback on the characteristics of the system, we vary the parameters β from 2 to 11, while the k_d value is fixed as 800.

The numerical results are shown in Fig. 1(a) for the κ_1 and κ_2 values and in Fig. 1(b) for the $\kappa_{1,2}^c$ and $\Delta \bar{G}$ values as functions of the equilibrium concentration of protein x_2^* for different β values. As a consequence of increasing the β value, the κ_1 and κ_2 values also increase but the $\kappa_{1,2}^c$ value decreases; the values are ranged between $0.305 \leq \kappa_1 \leq 0.354$, $-6.00 \times 10^{-4} \leq \kappa_2 \leq 6.26 \times 10^{-4}$, and $2.74 \times 10^{-4} \leq \kappa_{1,2}^c \leq 8.08 \times 10^{-4}$, respectively. Note that the κ_2 values are drastically smaller than the κ_1 values, reflecting the longer half-life of protein. Moreover, the $\Delta \bar{G}$ values for different β given in Fig. 1(b) indicate that the system becomes more in focus when the β value increases.

The fluctuations of the system near a stable point are analyzed by measuring the variance of x_i in terms of the Fano factors ν_i , defined as $\nu_i = \Omega (\sigma_{i,i}^2 / x_i^*)$, and the covariance of x_1 and x_2 in terms of the correlation coefficient R_{12} , defined as $R_{12} = \sigma_{1,2}^2 / \sqrt{\sigma_{1,1}^2 \sigma_{2,2}^2}$. The numerical results of ν_1 , ν_2 , and R_{12} for different β are shown in Fig. 2. The plots indicate that a larger κ_2 implies a smaller ν_2 and a larger $\kappa_{1,2}^c$ implies a larger R_{12} . Furthermore, as indicated in the inset of Fig. 2, the ν_1 values all are very closed to but less than one over the range of $2 \leq \beta \leq 11$; the value firstly decreases from $\nu_1 = 0.9844$ at $\beta = 2$, reaches the minimum $\nu_1 = 0.9827$ at $\beta = 4$, and then increases to $\nu_1 = 0.9926$ at $\beta = 11$. Because that the fluctuation of mRNA is caused by a process very close to Poissonian with $\nu_1 = 1$, we have very small correlation coefficient ranged between $0.015 \leq R_{12} \leq 0.101$. We further compare the results with those obtained from the linear noise Fokker-Planck equation which, as shown explicitly in Ref.[11], describes the distribution of fluctuations $\xi_i(t)$ introduced via the setting, $x_i(t) = \bar{x}_i(t) + \Omega^{-1/2} \xi_i(t)$, where the macroscopic values $\bar{x}_i(t)$ are determined by the rate equations of Eq. (1). The results thus obtained agree completely with those shown in Fig. 2.

In conclusion, we present a mechanical viewpoint on the characteristics of regulatory genetic networks, which is obtained from the energy landscape of the network near a stable point. The new approach is shown to be consistent with other descriptions, as demonstrated in the explicit calculations of auto-regulatory networks, it also provides additional informations, such as the steepness of a stable point and its relation to fluctuations. Though the method can also be applied to other genetic networks straightforwardly, it is limited in the sense that the overall potential landscape cannot be approximated by a local quadratic approach. For bi-stable systems such as toggle switches, one might need to patch the potential derived from the two local minimums in a rigorous fashion to obtain a better result.

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 - [16] The k_{\max} value was set as $2 \ln 2 \simeq 1.4$ in Ref. [8], this corresponds to a mean protein number 600 when $\beta = 0$ and leads to the increase of mean protein number as the Hill coefficient β increases. Our setting, $k_{\max} = 3$ which is the same as that given in Ref. [11], leads to the decrease of mean protein number as β increases. However, different settings do not change the mechanical picture obtained in this work.

FIG. 1: The force constants, κ_1 , κ_2 , and $\kappa_{1,2}^c$, and the effective free energy difference $\Delta\overline{G}$ for different β values: (a) κ_1 (solid squares) and κ_2 (hollow squares), (b) $\kappa_{1,2}^c$ (solid squares) and $\Delta\overline{G}$ (hollow squares). The horizontal axis is the equilibrium concentration of protein x_2^* .

FIG. 2: The Fano factors, ν_1 and ν_2 , and the correlation coefficient R_{12} for different β values. The horizontal axis is the equilibrium concentration of protein x_2^* , and the inset shows the details of the ν_1 values.



